Import risk analysis: Turkey rhinotracheitis virus in turkey hatching eggs from the United Kingdom sourced from TRT-vaccinated flocks

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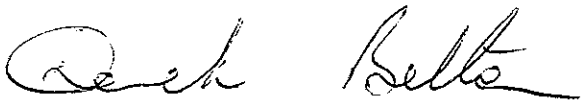

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EXECUTIVE SUMMARY

This document is an analysis of the risk of introducing turkey rhinotracheitis (TRT) virus into New Zealand from the United Kingdom in imported turkey hatching eggs obtained from parent stock vaccinated against that disease.

The need for this risk analysis arose due to the fact that vaccination for TRT, with both live and killed vaccines, is now common practice in all countries from which the New Zealand turkey industry can obtain the hatching eggs that form the basis of their breeding stock.

In this analysis the commodity definition is assumed to incorporate the current pre-export health requirements for parent flocks as well as the requirement for the application of OIE accepted egg sanitation practices, as set out in the current Import Health Standard (IHS) for turkey hatching eggs.

The introduction of TRT virus into New Zealand would result in a severe short-term disruption to the turkey industry due to respiratory disease and deaths of young turkeys, as well as a drop in egg production in laying birds. TRT virus would cause relatively mild losses if it were introduced into poultry species other than turkeys. Although there is serological evidence that some species of wild bird may be infected (see Appendix 1), there is no evidence that this organism can cause primary clinical disease in wild birds, and as such the risk to native birds is considered to be negligible. There are no public health risks associated with avian pneumovirus infections in poultry.

This risk analysis concludes that TRT virus does pose a hazard in turkey hatching eggs, but vaccines used to protect parent stock, be they attenuated or killed, do not pose a hazard. In view of the current pre-export requirements for fumigation and disinfection of hatching eggs prior to export it is considered that the likelihood of introduction of TRT virus into New Zealand in the commodity is very low. Further, in view of existing post-arrival quarantine requirements, likelihood risk of exposure is considered to be negligible.

Specific safeguards are recommended for the investigation of outbreaks of clinical respiratory disease in post-arrival quarantine and the ruling out of TRT as the cause.

INTRODUCTION

This document is an analysis of the risk of introducing turkey rhinotracheitis (TRT) virus into New Zealand in turkey hatching eggs imported from flocks in the United Kingdom that are vaccinated against the virus.

This risk analysis has been carried out by a private consultant, on behalf of the intended importer.

TRT virus is an avian pneumovirus (APV), which is now thought to be widespread in most countries with significant poultry production. Besides TRT in turkeys, APVs are associated with several disease syndromes in other poultry, including swollen head syndrome (SHS) and avian rhinotracheitis (ART) in broiler chickens (Gough, 2003).

The need for this risk analysis has arisen due to the fact that few turkey-producing countries now remain free from TRT and that the current accepted method of control includes the use of live and killed TRT vaccines to protect parent turkey breeder stock. In the recent past, the New Zealand turkey industry has been able to import turkey hatching eggs from Northern Ireland where TRT was not present, and where it was possible to find seronegative donor flocks that had not been vaccinated against the disease. However, with the recent occurrence of TRT in Northern Ireland, flocks from all suitable sources in the United Kingdom are now vaccinated against TRT, and in order to continue supply of eggs from the UK it would be necessary to source them from TRT vaccinated flocks.

COMMODITY DEFINITION

In this risk analysis the commodity definition is assumed to incorporate the current pre-export health requirements for parent flocks of imported turkey hatching eggs and the accepted OIE recommendations for egg sanitation practices, as set out in the current IHS which is included as Appendix 3 of this document.

Thus, the commodity is defined as "Turkey hatching eggs obtained from TRT vaccinated flocks in the United Kingdom". This means that it is taken as a given that the following specifications are also enforced:

- Parent flocks must be certified to be of acceptable health status and must not be vaccinated for Newcastle disease, as outlined in the certificate currently given as an example in the Import Health Standard for the import of turkey hatching eggs into New Zealand from Australia, Canada, England, Scotland, Wales and Northern Ireland.
- Parent flocks must test negative for *Mycoplasma gallisepticum*, *M. synoviae*, *M. meleagridis* and *M. iowae*, according to a MAF-approved protocol.
- Hatching eggs for export must comply with the sanitary conditions under Article 3.4.1.7 of the current International OIE International Animal Health Code.

- Imported hatching eggs, and the poults hatched from them, must comply with mandatory post arrival quarantine requirements currently in force[1], including serological testing for Newcastle disease after 6 weeks in quarantine.

The term 'turkey rhinotracheitis virus' has been used in this document to include all APV isolates which are primarily isolated from turkeys demonstrating signs of clinical disease attributable to TRT.

The term 'United Kingdom' is defined as including the countries of England, Scotland, Wales, and also Northern Ireland.

METHODOLOGY

The methodology used in this risk analysis follows the guidelines in Section 1.3 of the *International Animal Health Code of the Office International Des Epizooties* (OIE, 2002). In New Zealand, the OIE risk analysis framework is applied as described in *Import Risk Analysis Animals and Animal Products* (Murray, 2002).

The risk analysis process used by the MAF is shown in Figure 1.

Under the OIE methodology, for each potential hazard, the following analysis is carried out:

Risk Assessment

a) Release assessment - the likelihood of the organism being imported in the commodity.

b) Exposure assessment - the likelihood of animals or humans in New Zealand being exposed to the potential hazard.

c) Consequence assessment - the consequences of entry, establishment or spread of the organism.

d) Risk estimation - a conclusion on the risk posed by the organism based on the release, exposure and consequence assessments. If the risk estimate is non-negligible, then the organism is classified as a hazard.

[1] Under the existing IHS, the eggs must be hatched in a post-arrival quarantine facility under MAF Biosecurity Standard 154.02.05: Avian Transitional Facilities. The quarantine period will be long enough to assess the health of hatchlings and to carry out post-arrival testing and investigate any significant mortalities. Testing a random sample of 100 birds for avian paramyxovirus type 1 is carried out at 42 days of age.

Risk Management

a) Risk evaluation - a determination is made as to whether sanitary measures are necessary.

b) Option evaluation - identify the options available for managing the risk, and consider risk reduction effects.

c) Recommended measures - the recommendation of the appropriate option or combination of options that achieve a negligible likelihood of entry, spread or establishment, while minimising negative trade effects.

In this risk analysis, the Hazard Identification is confined to a consideration of TRT virus, which is defined as any strain of APV capable of causing TRT in turkeys, including field strains and vaccinal strains.

Therefore it is necessary to include an assessment of the potential risk associated with the use of live and killed vaccines for the control of TRT in parent turkey stock. This is with regard to

a) the potential of vaccination to mask clinical infection should adult turkeys be exposed to field strains of TRT virus, and

b) the risk of attenuated live TRT vaccine virus entering New Zealand in the commodity and becoming a hazard to poultry and other birds.

The release assessment considers the likelihood of introduction of field strains or vaccine strains of TRT virus in or on hatching eggs.

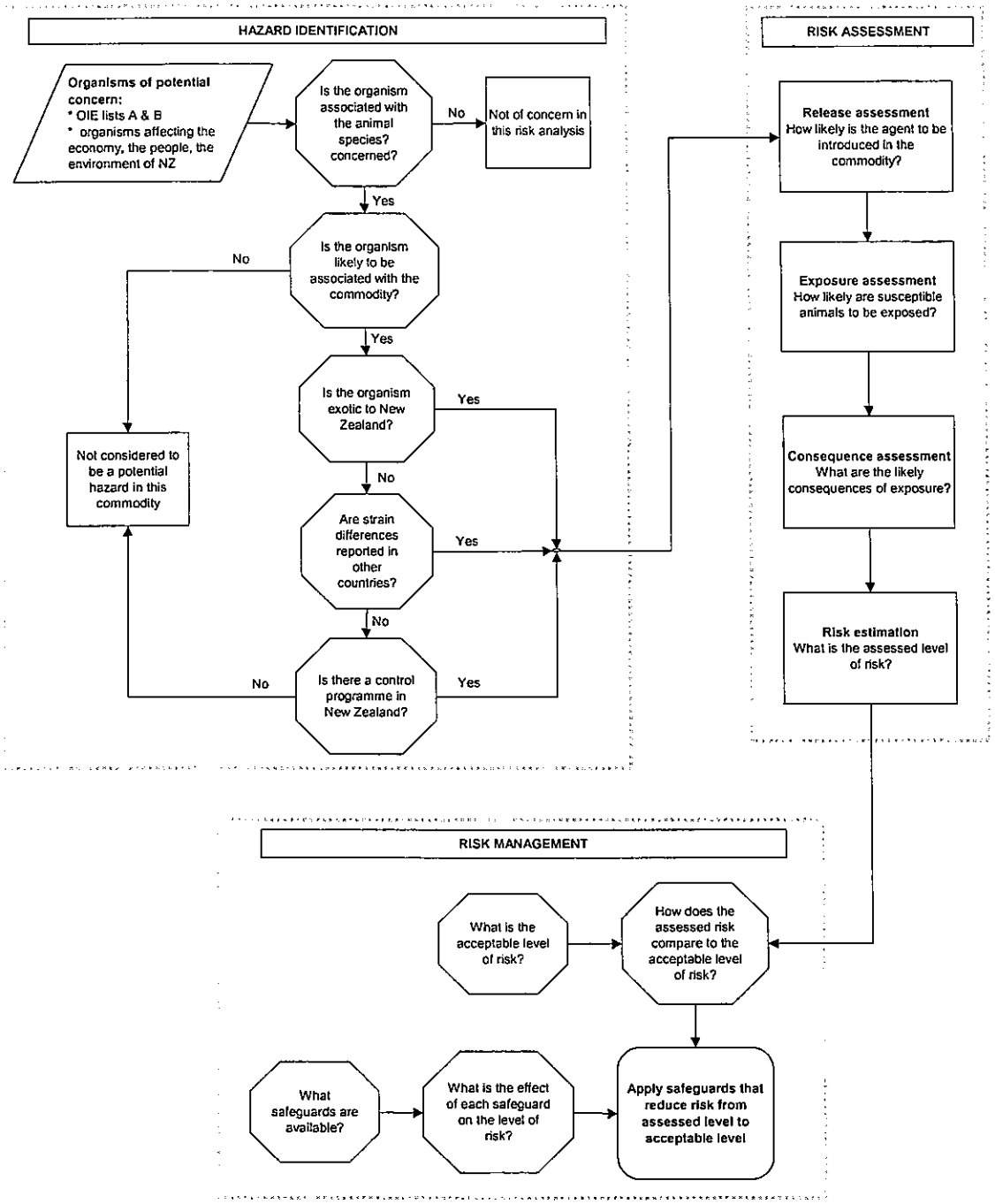
The exposure assessment considers the potential pathways by which susceptible species in New Zealand could be exposed to field or vaccinal strains of TRT virus, and the likelihood of such exposures occurring. However, since the commodity definition includes the requirement for post-arrival quarantine and testing for other disease agents and the requirement to investigate any outbreaks of clinical disease that occur in quarantine[2], the exposure assessment will be limited to the likelihood of the virus escaping from post-arrival quarantine given the existing requirements.

The consequence assessment will examine the likely consequences of TRT virus exposure to susceptible species, if it were to escape from post-arrival quarantine.

The risk management section will consider what additional safeguards, if any, need to be added to the current post-arrival quarantine standard to manage the risk of TRT virus.

[2] MAF Biosecurity Standard 154.02.05: Avian Transitional Facilities, especially section 4.2.4 'Supervision'

Figure 1. The risk analysis process.



1. HAZARD IDENTIFICATION

1.1 AGENT

Turkey rhinotracheitis (TRT) is caused by a single-stranded, enveloped, non-segmented RNA virus belonging to the genus *Metapneumovirus,* in the subfamily *Pneumovirinae,* within the family *Paramyxoviridae* (Pringle, 1998).

The literature on avian pneumoviruses and on the clinical syndrome of TRT is very broad, but the terminology used varies, with some authors referring to the causative agent as 'turkey rhinotracheitis virus' and others using the more general term 'avian pneumovirus' or APV.

1.2 OIE LIST

TRT virus is not listed by the OIE.

1.3 NEW ZEALAND'S STATUS

TRT virus is exotic to New Zealand, and is listed on the unwanted organisms register.

The clinical syndrome of turkey rhinotracheitis has not been reported in turkeys in New Zealand and limited serological work suggests that New Zealand poultry are free of infection with avian pneumoviruses (Horner, 1993).

1.4 EPIDEMIOLOGY

The disease

APV was first identified in disease outbreaks in turkeys in South Africa in the 1970s (Buys and du Preez,1980; Buys et al, 1989), after which it appeared in turkey flocks in Europe and in the United States of America (Naylor and Jones, 1993).TRT was first seen in the United Kingdom (UK) in 1985 in East Anglia (Alexander et al, 1986), and within 6 months it had spread throughout England.

TRT can result in significant economic and welfare problems in susceptible populations of turkeys (Cook, 2000), but there is uncertainty about the relative role of concurrent disease and secondary bacterial invaders, such as *Bordetella avium*, *Ornithobacterium rhinotracheale* and the *Mycoplasma* group of microbes, and of management practices (Gough, 2003).

In susceptible turkey flocks, TRT is characterized by a very high morbidity, approaching 100% in young birds, with clinical signs of acute upper respiratory disease (Stuart, 1989). Mortality rates, following severe respiratory distress, are very variable, ranging from very low levels to as high as 50% (Pattison,1998; Stuart, 1989). This variation is probably due

to different management practices, with factors such as poor ventilation, high stocking rates and mixed age sheds being associated with higher mortality rates. Concurrent disease alters the clinical presentation and the outcome of infection. Husbandry practices such as debeaking poults, where this occurs before or during the infection, can also increase the susceptibility of young turkeys to the development of clinical disease.

In laying turkeys infection with TRT virus generally results in a much milder respiratory infection (Stuart, 1989), but may result in a significant decline in egg production. Production losses of up to 60% have been reported in turkey layers and recovery can take 2-3 weeks. Fertility may not be affected, but where egg quality is impaired, the poults hatched from those eggs tend to be of poor quality (Stuart, 1989).

Infections in birds other than turkeys

Infection of chickens with APV causes SHS (Shin et al, 2000a, 2000b), which is characterized by the swelling of infraorbital and periorbital sinuses. In severe cases torticollis, cerebral disorientation and opisthotonus may occur although usually less than 4% of the flock become affected in this way. However, mild respiratory disease, associated with APV, may be more widespread (Gough, 2003). Mortality rarely exceeds 2% but egg production and egg quality may be adversely affected (Gough, 2003).

It is thought that severe clinical disease is due to secondary infection with *Escherichia coli* (Gough, 2003), and that this is more common in broiler breeders than in other classes of poultry. Clinical disease has been reproduced experimentally by intranasal inoculation of turkey isolates of APV in guinea fowl and pheasants (Gough, 2003).

Although antibodies to APV have been found in seagulls in the Baltic (Heffels-Redman et al, 1998; Cook, 2000) and in an ostriches in Zimbabwe (Cadman et al, 1994) there is no evidence to suggest that other avian species develop clinical disease as a result of infection with TRT virus. In one study, a range of poultry, including ducks, geese and pigeons, were given an intranasal inoculation of an infective dose ($10^{4.5}$ $TCID_{50}$) of a turkey isolate of APV. This resulted in mild disease in turkeys and conjunctivitis in pheasants, but failed to induce clinical disease in the other species. There was no measurable antibody response detected in the ducks, geese or pigeons (Gough et al, 1988), which led Cook (2000) to the conclusion that ducks, geese and pigeons may be refractory to infection with the APV isolate used. It is likely that the lower population density and other epidemiological factors make it less likely for wild birds to become exposed to the conditions required to potentiate APV associated disease. Although there is no direct evidence to implicate wild birds in spreading the virus from infected turkey flocks to other areas, this possibility cannot be dismissed completely (Cook, 2000).

Pathogenesis

The understanding of the pathogenesis of TRT and the nature of the causative agent has progressed significantly over recent years as a result of several research projects (Cook et al, 1991; Cook et al, 1993) and the development of reliable diagnostic tests (Cavanagh et al, 1997; Mekkes and de Witt, 1998). Advances in molecular biology have also lead to better characterisation of the APV subtypes that are involved in TRT (Seal, 2000).

The incubation period for TRT in turkeys is short – often only a few days in the field situation. However, the development of clinical signs can depend on other factors (Cook, 2000). In experimental infection of turkeys, TRT virus can be detected in the nasal area, the trachea and the airsacs within one day of inoculation, and can persist for approximately ten days (Cook, 2000; Jones, 1996). Clinical signs of TRT develop from the third day post inoculation and abate by day nine (Cook, 2000). This relates well to the 6-7 day period of virus shedding (days 3 or 4 to 9 or 10 post intranasal inoculation) reported by Jones (1996). Current evidence suggests that a carrier state does not exist in turkeys and that the virus is not latent in recovered birds (Cook, 2000)

Transmission

Since avian pneumoviruses infect the upper respiratory tract and cause acute respiratory signs in infected birds, aerosol transmission was considered likely (Cook, 2000). However, in the UK outbreak it is thought that the rapid spread between turkey flocks and farms was due to movement of affected or recovered poultry. Transmission of the disease via contaminated water and movement of personnel and equipment has also been suggested. As stated earlier, the role of wild birds remains the subject of speculation (Cook, 2000).

Strain variation

In the United Kingdom and Europe, one serotype of APV is recognized on the basis of cross neutralization studies, but it contains two subtypes (A and B) based on the molecular sequencing of the G glycoprotein (Juhasz and Easton, 1994) and confirmed by monoclonal antibody studies (Cook, 2000). In the United Kingdom, both subtypes A and B may cause disease in experimentally infected turkeys and chickens but the disease in turkeys seems to be more severe (Cook, 2000). APV subtypes A and B have also been reported in turkeys farmed in various countries in Europe (Toquin et al, 1999).

In the USA isolates of APV from Minnesota and Colorado (designated as subtype C or the Colorado 'strain') appear to be of a different serotype (Seal ,1998), which does not show cross neutralisation with the UK isolates (Gough, 2003). This initially caused some difficulty in the serological diagnosis of the disease in the USA (Seal, 2000). This subtype has only been reported in turkeys in the USA (Gough, 2003).

A new type of APV recently reported in Muscovy ducks in France appears to be more closely related to the Colorado 'strain' than to the A or B subtypes of APV typically identified in Europe but it is antigenically different from the USA isolates (Toquin et al, 2000).

Although there appears to be differing species tropism of different APV strains, there is no evidence of 'strain' differences resulting in variation in incubation period or clinical signs in turkeys (Cook, 2000). Notwithstanding this, in experimental infections of chickens, chicken isolates of APV cause more severe clinical signs than do turkey isolates (Cook, 2000).

It is likely that there are more strains of APV than have been identified to date but that their role in avian disease may only be secondary.

Vaccination Practices in UK breeder turkeys

Vaccination against TRT is commonly practised for both turkey and broiler breeder flocks and this has been successful in reducing morbidity associated with TRT in turkeys and also in reducing losses due to SHS or ART in chickens.

Breeder turkeys in the United Kingdom are vaccinated with live and killed TRT vaccines. Vaccination practices change in response to new developments in vaccine research and may also reflect changes in the epidemiological profile of disease. The principles of vaccination are well explained in text books on poultry husbandry and poultry diseases (e.g. Cserep, 2001). A typical programme for the vaccination of UK breeder flocks against TRT is shown in Table 1.

Table 1: A typical vaccination programme for turkey breeders (Cserep, 2001).

Age for innoculation	Type of vaccine	Formulation of vaccine
Day one	TRT live **	Coarse spray
Week 6	TRT live	Coarse spray
Week 10 *	TRT live	Coarse spray
Week 14	TRT killed ***	Injection
Week 22-24	TRT killed	Injection

Explanatory notes for Table 1:

* The vaccine at week 10 is optional

** Live (attenuated) vaccines delivered by coarse spray (or as an eye drop) are effectively used to stimulate local immunity in the respiratory tract of the young birds. Several doses are given to provide sufficient stimulation to facilitate recognition of the foreign agent by the bird's local and cell mediated immune system. Attenuated strains of field virus have been used successfully to protect turkey parent stock against TRT and have been well studied in research trials (Cook et al, 1989, 1995, 1996).

*** Killed vaccines are given to further stimulate the immune system and to promote the development of protective antibodies. These antibodies may be passed in the egg to the chick but do not give protection against TRT for the hatched poults (Naylor et al, 1997).

1.5 HAZARD IDENTIFICATION CONCLUSION

Since TRT virus is not present in New Zealand, it is considered to be a potential hazard in this commodity.

2. RISK ASSESSMENT

2.1 RELEASE ASSESSMENT

This release assessment will consider the likelihood of field or vaccinal strains of TRT virus being introduced with imported turkey hatching eggs in two ways, by making a distinction between carriage of the virus within the egg (by true vertical transmission) and transmission on the surface of the egg (by contamination of the egg shell with the virus via secretions or excretions of infected turkey layers).

2.1.1 Carriage in eggs

Unvaccinated birds

Although there is no evidence that vertical transmission of avian pneumovirus occurs in field outbreaks (Khehra and Jones, 1999; Jones et al, 1988), the virus has been demonstrated in oviduct tissue or in the oviduct of infected parent stock. Further, egg production can be severely compromised by TRT infections in adult turkey hens, although the effect of APV infection on egg production in broiler breeders is considered to be less pronounced.

Jones et al (1988) demonstrated that when serologically negative breeding turkeys were given an intranasal inoculation of TRT virus, it was possible to find the virus in the oviduct at days 7 and 9 post-inoculation (p.i.). Evidence of the presence of viral antigen was detected (using immunofluorescence) in the uterus on day 7 p.i. and in the oviduct and vagina on day 9 p.i. using tracheal organ culture, virus was also isolated from the middle magnum and vaginal tissues, on day 9 p.i. However, antigen was not found in any tissue samples collected on day 12. On other occasions, all tissues were negative for virus up to 20 days p.i. Antibodies reached a high titre by 12 days p.i and were maintained at a high level ($\log_2$ 12-15) throughout the period of observation (89 days). The authors comment that this work was carried out in serologically negative birds, and that different results might occur in serologically positive birds (Pattison, 1998) – vaccines were not commercially available until 1989.

These findings suggest at least a theoretical possibility that APV could be present in eggs for a short period during an outbreak of TRT in a susceptible laying flock. Although vertical transmission does not seem to be a significant method of transmission for TRT virus in field outbreaks, further work appears necessary to confirm this. In considering this possibility, Cook (2000) concluded that although viral antigen can be detected in the reproductive tract of infected laying birds, it was not clear if the oviduct tissue was actually susceptible to APV infection. While in vivo studies using intravenous inoculation of laying birds failed to demonstrate virus replication in the oviduct, concurrent *in vitro* studies illustrated the intrinsic susceptibility of the oviduct tissue in chickens (Khehra and Jones, 1999). The significance of these findings is unclear, but it does indicate lack of evidence to rule out completely the possibility of vertical transmission of TRT virus. Moreover, TRT virus has been isolated from neonatal turkey poults (Shin et al, 2002).

Vaccinated birds

In view of the importance of adequate levels of circulating antibody to protect the oviduct against viral replication (Jones, 1996) the use of TRT vaccines in layer birds is recommended to produce high levels of circulating antibody (Baxendale, 1996).

When vaccinated turkeys were experimentally challenged with TRT virus at 38 weeks of age it was noted that there was significant protection against the TRT-associated drop in egg production (Cook et al, 1996). This was achieved using a single dose of live attenuated vaccine administered at 1 week of age. When a combination of live (attenuated) and a killed vaccine was used, experimentally challenged birds were protected against respiratory signs of TRT as well as the associated drop in egg production when compared to unvaccinated controls (Cook et al,1996).

These results and the lack of virus excretion from vaccinated turkey hens following virus challenge, support the view that the eggs of healthy TRT vaccinated turkeys are unlikely to carry field strains or vaccinal strains of TRT virus (Cook, 2000).

In view of the above, it is generally accepted that a modern comprehensive vaccination programme (using a combination of live and killed vaccines, such as that shown in Table 1) will reduce the risk of virus excretion in TRT infected birds and also prevent TRT field challenge virus being able to multiply in the oviduct of laying birds.

2.1.2 Carriage on eggs

Although the virus has been isolated from the trachea, lung and viscera of infected turkey poults, by far the best source of virus has been the upper respiratory tract. While this finding could imply that aerosol transmission is the most likely route of spread between birds, this has not been demonstrated in field situations (Cook, 2000). In addition, although it is theoretically possible for contamination of the shell to occur during egg formation (as discussed above for the egg contents), faecal shedding of TRT virus is thought to be insignificant (Jones, 2001).

Since TRT virus is an enveloped RNA virus, it is unlikely to survive for very long outside the host (Jones 1996). The virus survives only 2 days at 37°C (Gough, 2003). It is susceptible to a range of disinfectants and is considered to be readily inactivated by routine egg sanitation practices (Cook, 2000). This view is consistent with field experience where strict attention to hygiene and the enforcement of biosecurity measures has been successful in reducing the spread of the disease (Stuart,1989; Pattison, 1998).

Thus, although eggs from infected birds could theoretically be contaminated by respiratory secretions or faecal material from adult turkeys, this has no significance in terms of disease transmission in the field. In view of these findings, it is considered that the likelihood of TRT virus contaminating eggs is extremely low, even during an outbreak. Cook (2000) considers that, providing elementary egg hygiene and disinfection processes are followed, there is a negligible risk of importing TRT virus on hatching eggs.

2.1.3 Release assessment conclusion

Although there is no direct evidence to suggest that turkey rhinotracheitis virus is passed vertically from infected parent stock to the egg, particularly in vaccinated birds where infection of the oviduct is minimised, it cannot be concluded that the likelihood of this is negligible.

The likelihood that TRT virus will be carried on the surface of eggs is very low. Given the requirement for the collection of eggs from healthy parent stock, the requirement for egg sanitation practices to be applied, and the fragile nature of the virus, the likelihood of TRT virus being present on the surface of eggs laid by vaccinated turkeys is negligible.

Therefore, it is concluded that there is a non-negligible likelihood that eggs imported from TRT vaccinated breeder turkeys in the UK would carry TRT virus.

2.2 EXPOSURE ASSESSMENT

Direct contact is thought to be required for the transmission of TRT to occur between infected and susceptible turkeys. Transmission to turkey poults in separate pens in the same airspace did not occur under experimental conditions (Cook, 2000). Contaminated water, movement of affected or recovered poults, and fomites have been implicated as methods of spread in field outbreaks (Cook, 2000). While airborne spread and vertical transmission have been suggested as possibilities, at present only contact spread has been confirmed (Gough, 2003).

Spread of avian pneumoviruses to other birds from turkeys is possible by any of the routes outlined but there is no direct evidence to demonstrate whether or not this occurs in field situations. Moreover, as is discussed in Appendix 1, although there is some speculation about possible spread by wild birds, this has not been proven.

During the initial stages of the TRT outbreak in the United Kingdom in 1986 it became clear that flocks at all levels of the breeding pyramid were infected almost simultaneously (Pattison, 1998). It appeared impossible to prevent the rapid spread of the agent to susceptible populations of turkeys in commercial turkey farms and most breeder farms (Stuart, 1989). There was a slower rate of disease spread in Scotland than in England, apparently due to lower stocking densities (Cook, 2000).

In New Zealand, even in North Canterbury, the turkey population density is even lower than in Scotland. Nevertheless, the epidemiology of TRT, and the lack of any immunity to TRT in the New Zealand turkey population suggests that if the virus were to be introduced then its transmission to turkey establishments and broiler operations would occur. This would most likely occur by direct or indirect contact i.e. feed lorries, staff, and from farm to farm by the movement of birds that had recovered from the disease.

Since clinical signs of TRT infection are not prevented by the presence of maternal antibodies in turkey poults (Naylor et al, 1997), an outbreak of the disease in quarantined

turkey poults is theoretically possible if TRT virus were present in hatching eggs from TRT vaccinated stock. However, the current post arrival quarantine (PAQ) conditions imposed by 154.05.02 (Standards for Avian Transitional Facilities) are designed to contain avian paramyxovirus type 1 (Newcastle disease virus) if it were introduced with imported hatching eggs. Thus, the likelihood of birds outside the quarantine building being exposed to TRT, even if a serious outbreak of TRT were to occur in the quarantined birds, is negligible

Moreover, the quarantine standard requires that any unusual clinical disease in quarantined birds is investigated by MAF, and the very obvious clinical signs of TRT that would be expected if the virus were present in quarantined birds means that detection would be certain.

2.3 CONSEQUENCE ASSESSMENT

The consequences for the turkey industry, if release and exposure to TRT virus were to occur, would most likely be similar to that seen in other parts of the world, that is, significant mortality in young turkeys and poor egg production in adult turkeys (Gough, 2003). These initial losses in susceptible birds would be brought under control by the use of vaccination. The effect would be significant economic losses in the short term and ongoing control costs for the turkey industry in the longer term. The USA experience suggests that once TRT appears in the turkey population in an area, eradication is extremely difficult to achieve (Harvorson, 2003).

In most cases where chickens are infected with turkey isolates of APV, concurrent bacterial infection is necessary for severe clinical disease to occur (Hafez, 1993), so the consequences for the broiler industry would probably be less serious than the consequences for turkeys, and the losses would be controllable by good husbandry practices. Although vaccination against turkey isolates of APV has been found to be beneficial for broiler breeders in the UK (Pattison, 1998), where this is now standard practice, vaccination of commercial broilers is not generally cost-effective. Nevertheless, the impact for this and other sectors of the poultry industry in this country in the event that TRT virus were introduced could be severe.

In the case of non-gallinaceous birds, whilst there are reports of detection of antibodies to APV in different avian species in Europe, Africa and USA, and isolation of APV from ducks in France, there is no evidence of any associated disease. The risks to native birds are unknown, but these may be expected to be low due to the fact that TRT, ART and SHS are diseases of farmed gallinaceous birds, particularly under conditions of high stocking density and poor husbandry (Stuart, 1989; Pattison, 1998).

There are thought to be no public health risks associated with APV infections in poultry (Gough, 2003).

2.4 RISK ESTIMATION

The likelihood of vaccinal or field strains of TRT virus being present in turkey hatching eggs is considered to be very low, particularly in vaccinated birds. In view of the current requirement to clean and disinfect turkey hatching eggs prior to export and the requirement for appropriate health certification, it is considered that the risk of release of this hazard in to New Zealand, in the defined commodity, is very low. The likelihood of exposure is considered to be negligible in view of the post-arrival quarantine requirements that are currently in place for imported hatching eggs. However, since there would be significant consequences for the poultry industry if TRT virus were to be introduced, it can be concluded that the risk is non-negligible.

3. RISK MANAGEMENT

3.1 RISK EVALUATION

In view of the non-negligible risk posed by TRT virus in imported turkey hatching eggs, safeguards are required to manage the risk in post-arrival quarantine.

3.2 OPTION EVALUATION

3.2.1 Risk management objective

The risk management objective is to prevent the release of TRT virus from post-arrival quarantine in New Zealand.

3.2.2 Options available

3.2.2.1 Serological testing of hatchlings in quarantine

Serological tests could be used to detect antibodies in turkey poults that are hatched in quarantine, and several commercial ELISA kits are available that could be used for this purpose (Eterradossi et al, 1995; McFarlane-Toms et al, 1998; Mekkes and de Wit, 1999; Toquin et al, 1996).Sampling protocols can be designed to give high levels of statistical confidence of detecting TRT antibodies at a stated threshold prevalence. However, many of these tests have been developed for the purpose of demonstrating the level of antibody in vaccinated hens (hence the level of protection provided by the vaccine) rather than for the diagnosis of TRT infection itself, and their diagnostic sensitivity and specificity is not well established.

A further difficulty in relying on antibody tests in hatched poults is that even if the poults were tested 6 weeks after hatching, it is possible that a proportion of birds could test positive as a result of residual levels of maternal antibody being present. Although this is considered unlikely (Gough, personal communication)[3] any positive tests would require follow-up testing by other methods to exclude infection with TRT virus.

3.2.2.2 Investigation of clinical disease in quarantine

Although it is implicit in the MAF quarantine standard that any unusual mortalities and clinical disease in birds in quarantine must be investigated, a case can be made for the development of specific requirements for TRT virus testing of turkey poults that are hatched in quarantine and which display signs of respiratory disease.

In view of the highly infectious nature of TRT virus, if it were present in post-arrival quarantine it would spread very rapidly to infect close to 100% of birds, and in view of the very obvious clinical signs in immunologically naïve birds, regardless of residual

[3] Richard E Gough, VLA Weybridge, UK. External critique of this risk analysis, dated 16 December 2003.

maternal antibody, infection would be very obvious. Testing of such birds, by virus isolation using tracheal organ culture and/or RT PCR testing for TRT virus would be appropriate.

3.3 RECOMMENDED MEASURES

It is recommended that the following measures be added to the current IHS.

- parent turkey stock, from which turkey hatching eggs are obtained, have no clinical signs of turkey rhinotracheitis and that there have not been any outbreaks of TRT in the area around the turkey breeding establishment during the immediate period prior to, or during, egg collection.

- in case of any outbreak of respiratory disease in quarantine, affected birds must be tested for TRT virus by virus isolation using tracheal organ culture and/or PCR.

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APPENDIX 1

AVIAN PNEUMOVIRUS INFECTION IN NON-GALLINACEOUS BIRDS

Stuart (1989) first suggested that infection of non-gallinaceous migratory birds might have been responsible for the introduction of TRT virus to the UK, solely on the grounds that the disease first appeared in East Anglia, which is the first UK landfall for many of these birds. The monoclonal antibody analyses of Cook et al (1993) that demonstrated similarities between a 1978 APV isolate from South Africa and a 1985 UK isolate have been quoted as supporting evidence, but there has been little additional evidence to back this up. In their review, Naylor and Jones (1993) indicated that there had been no reports of spread by wild birds. Others consider that pigeons, geese and ducks are refractory to the virus (Gough et al, 1988), although APV has been isolated from farm-reared pheasants in the UK with an acute upper respiratory tract infection (Gough et al, 2001). Evidence against transmission of the disease by wild birds includes the fact that Northern Ireland remained free of APV until 1996, despite its proximity to infected parts of the UK (Jones 1996).

However, antibodies to APV have been demonstrated in ostriches in Zimbabwe (Cadman et al 1994) and sea gulls (Heffels Redmann at al 1998) in the Baltic. Moreover, researchers in Minnesota (Shin et al, 2000) working with the USA strain of APV were able to demonstrate the presence of APV RNA and antibodies to APV in wild birds (geese, sparrows and starlings). The viral genome showed a close genetic identity with virus circulating in neighbouring turkey flocks, leading to the authors' conclusion of a common source. Moreover, Shin et al(2002) later succeeded in isolating infectious APV from sentinel ducks placed next to an APV-infected turkey farm. These findings suggest that wild birds may indeed act as reservoirs of APV and that this may explain the periodic pattern (March-April, October- November) of APV outbreaks in the USA that have not been attributable to management practices. However, the same authors point out that it is difficult to attribute the spread of APV within the north-central states to bird migration alone, since Canada to the north and US states in the south (which are on migratory fly-ways) have not reported APV outbreaks. Gough (2003) comments on the lack of spread of the USA strain of APV from Minnesota, despite the significant migratory bird population, and also on the lack of spread of this strain to chickens.

Therefore, although there is some evidence that ducks can become infected with the USA strain of APV, Gough's (2003) summary suggests that this does not happen with European strains.

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APPENDIX 2

IMPORT HEALTH STANDARD FOR THE IMPORTATION OF TURKEY HATCHING EGGS INTO NEW ZEALAND FROM AUSTRALIA, CANADA, ENGLAND, SCOTLAND, WALES AND NORTHERN IRELAND

Issued pursuant to Section 22 of the Biosecurity Act 1993
Dated: 5 September 2002

1. PERMIT

1.1 A permit to import must be obtained from Import Management, Ministry of Agriculture and Forestry (MAF), P O Box 2526, Wellington, New Zealand.

1.2 Permits will be issued for single consignments only, and the importer must supply the following information:

1.2.1 name and address of exporter
1.2.2 breed and type of poultry
1.2.3 number of eggs to be imported.
1.2.4 date of the proposed importation
1.2.5 location of the nominated avian transitional facility in New Zealand
1.2.6 a letter from the Veterinary Officer supervising post-arrival quarantine stating that:

- the facility meets the facility approval requirements of *MAFBA Standard 154.02.05 Standard for Avian Transitional Facilities* and is able to accept the proposed number of eggs to be imported on the proposed date of importation.
- the method and route of transport has been approved from the port of arrival in New Zealand to the approved avian transitional facility.

2. DOCUMENTATION

The permit and all the required certification must accompany the consignment to New Zealand.

3. ELIGIBILITY

3.1 The birds from which the eggs have been obtained must have been hatched in or have been in the exporting country for at least six months and during this time have not had any contact with imported birds.
3.2 The eggs have to be clean and have been fumigated.

3.3 The eggs have to be placed and sealed in gas impermeable plastic bags and sealed into clean and disinfected crates, using an official seal, attached by a full-time government veterinary officer of the country of origin before despatch.

4. IDENTIFICATION

The consignment of eggs must be clearly identified and identifiable with the health certification.

5. QUARANTINE

The eggs shall be hatched in New Zealand in an approved transitional facility complying with Std 154.02.06: Standard for Avian Transitional Facilities, and the hatched birds shall remain in this facility for at least 60 days.

6. ENTRY CONDITIONS

6.1 Details of transport and arrival times must be supplied to the Port Veterinary Officer at the airport of entry not less than 7 days in advance of importation.

6.2 On arrival in New Zealand the consignment will be checked by an Inspector under the Biosecurity Act 1993 and, provided the documentation is in order and seals intact, the eggs will be directed to the transitional facility nominated in the permit.

6.3 Vehicles whilst being used to transport crates to the transitional facility must not transport any other eggs or poultry.
The vehicles must be thoroughly cleaned and disinfected after delivery of the eggs.

6.4 After the eggs have been placed in an incubator all fillers etc and packing must be destroyed by incineration. The crates may be used again if they are fumigated.

6.5 All birds hatched in the transitional facility will remain in the facility for a period of not less than 60 days.

6.6 During that time any unusual deaths and/or sicknesses must be reported to the nearest MAF veterinary officer and carcasses retained for possible post-mortem examination.

6.7 MAF reserves the right to remove birds and/or specimens for each tests as may be desired at any time.

6.8 Blood samples are to be taken for serological testing for Newcastle Disease at the National Centre for Disease Investigation, Wallaceville when the birds are six weeks old. Samples should be taken from 15 birds or 10% whichever is smaller.

6.9 Regular inspections will be made of the farm by the veterinary supervisor.

6.10 No compensation will be paid for birds slaughtered as a result of test for disease or for diagnosis.

VETERINARY CERTIFICATE:

Species: TURKEY HATCHING EGGS
To: NEW ZEALAND
Import Permit Number .....

Exporting Country:
Ministry/Department:
Region:

I: IDENTIFICATION OF EGGS

Number	Species	Identification

Total Number: .....

II: SOURCE OF EGGS

Name and address of exporter: .....
.....
Name and address of breeder: .....
.....

III: DESTINATION OF EGGS

Name and address of consignee: .....
.....
Means of transport: .....

IV: SANITARY INFORMATION

OWNER'S/MANAGER'S DECLARATION

I, ... being the owner/manager of the flock from which the eggs have originated certify that:

1 The birds in the flocks from which the eggs are obtained have either been hatched in or have been in the exporting country for at least six months and during this time they have not had any contact with imported birds.

2 After collection only clean eggs have been selected and fumigated with formaldehyde using one of the methods in Article 3.4.1.7 of the OIE *International Animal Health Code*, namely formaldehyde gas using:

either

2.1 Method 1: ....ml formalin (....%) and ....g potassium permanganate per cubic metre of fumigation space.

or

2.2 Method 2: evaporation of 10g of paraformaldehyde powder or pellet per cubic metre of fumigation space.

(*Delete 2.1 or 2.2 as applicable.*)

3 There has been no clinical evidence of Lymphoproliferative disease of turkeys (LPDT) during rearing in the flock of origin.

(Owner/Manager)

Date

VETERINARY CERTIFICATE

I, ... being a Veterinary Officer employed by the government of.....(exporting country) certify with respect to the eggs identified in this certificate that:

1 OWNER'S DECLARATION

After due enquiry I have no reason to doubt the owner's/manager's declaration.

2 AREA FREEDOM

Highly Pathogenic Avian Influenza, Newcastle Disease and turkey rhino tracheitis has not occurred on any premises within a radius of 25 km of the premises of origin for a period of six months.

3 FLOCK OF ORIGIN

3.1 During the 30 days prior to the collection of the eggs, the birds in the flock of origin were inspected and found to be free from clinical evidence of infectious diseases including Newcastle disease, avian influenza, avian leucosis , avian reticuloendotheliosis, big liver and spleen disease, egg drop syndrome, fowl pox, infectious avian encephalomyelitis, infectious bronchitis, infectious bursal disease, infectious laryngotracheitis, mycoplasmosis, ornithosis, paramyxoviruses 2, 3 and 7, pasteurellosis (acute fowl cholera), salmonellosis, tuberculosis, turkey haemorrhagic enteritis and vibronic hepatitis. To the best of my knowledge and belief, and after due enquiry none of these diseases have existed within the flock of origin during the preceding six months.

3.2 To the best of my knowledge and belief no live Newcastle disease vaccine or any other live vaccines have been used on the supply flock at any time during the three months before collection of the eggs.

3.3 All flocks from which the eggs were obtained are certified free of *Salmonella pullorum* and *S. gallinarum*.

4 TESTING

4.1 Within 30 days before the collection of the eggs for shipment, the birds in:

4.1.1 unvaccinated supply flocks have been subjected to a serological test to confirm with at least 99% confidence of detecting a prevalence of 5%, that the flock is seronegative to avian paramyxovirus 1 (APMV-1).

4.1.2 vaccinated supply flocks have been subjected to virus isolation from cloacal swabs to demonstrate with at least 99% confidence of detecting a prevalence of 5%, that there are no APMV-1 viruses circulating in the flock.

4.2 Within 30 days before the collection of the eggs for shipment, 10% of the birds in the supply flock have been subjected with negative results to:

4.2.1 The slide agglutination test or the haemagglutination test for mycoplasma (*M. iowae* and *M. melegridis*) (100% of the birds must be tested for *M. meleagridis* with negative results).

4.2.2 The complement fixation test or agar gel precipitation test or haemmaglutination inhibition test for influenza A virus.

4.2.3 ELISA test for turkey rhino tracheitis virus.

5 CONTAINERS

The eggs have been placed and sealed in gas impermeable plastic bags and sealed into clean and disinfected crates using an official seal.

Veterinary Officer employed by the

Official stamp and date

Government of .....

(exporting country)